MiniPrep

NucleoSpin® Plasmid DNA purification Kit

1. Preparation
   a. Make sure that RnaseA has been added into Buffer A1
   b. Make sure that ethno has been added into Wash Solution (stored at 4°C)
   c. Make sure that A2 and A3 don't have any sediment
2. Extract 1.5-5ml overnight suspension culture and centrifuge at 11,000 x g for 1 minutes to recollect bacteria and discard culture.
3. Add 250µl BufferA1 and suspend bacteria
4. Add 250µl BufferA2, immediately overturn the tube for 6-8 times. Stay in room temperature for 2-5 minutes to split bacteria.
5. Add 300µl BufferA3. Large amount of flocks appear. Overturn the tube for 6-8 times. Be careful don't let the flocks disperse.
6. Centrifuge at 11,000g for 6-10 minutes. Move supernatant into an NucleoSpin® Plasmid/Plasmid(NoLid) column and centrifuge 11,000 x g for 1 min. Discard liquid in collection tube.
7. (Optional) Add 500µl Buffer AW and centrifuge 1 min at 11,000 x g. Discard liquid in collection tube.
8. Add 600µl BufferA4, centrifuge at 11,000 x g for 1 min. Discard liquid in collection tube.
9. Centrifuge empty tube at 11,000 x g for 2min

(Using a vacuum centrifuge enrichment machines concentration, using a vacuum centrifuge enrichment machines concentration of alcohol solvent model 45 degrees 3 minutes, you can effectively remove the residual alcohol, to ensure the quality of plasmid elution.).

1. Add 50 µl Buffer AE, incubate under room temperature 1min, centrifuge at 11,000 x g for 1 min to elute DNA.